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Postharvest Salicylic Acid and Melatonin Dipping Delay Ripening and Improve Quality of 'Sensation' Mangoes

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Freshly harvested mature-green 'Sensation' mangoes were dipped for 10 min in 2 mM salicylic acid (SA) and 0.2 mM melatonin (MT) alone or in combination and stored at 23±1°C and 60–70%RH. SA or MT delayed ripening measured as higher green peel color (lower a* values), firmness, titratable acidity (TA) and membrane stability index (MSI) and lower weight loss, total soluble solids (TSS), TSS/TA ratio and hydrolytic enzyme activities (polygalacturonase, xylanase and α-amylase) compared to that of untreated fruit. Total phenolic content (TPC) in peel of treated fruit increased with fluctuations during ripening and was higher than the control. In pulp, TPC decreased up to 6 days but increased thereafter, and was higher in treated fruit than the control. Total flavonoid content (TFC) in the peel decreased while that in the pulp increased with fluctuations during ripening and was higher in treated fruit than the control. Vitamin C content decreased during ripening and was not affected by the treatments. DPPH radical scavenging capacity (RSC) in peel decreased during ripening and was higher in treated fruit than the control. However, RSC in pulp increased during ripening and was not affected by treatments, except for SA plus MT treatment that gave higher RSC than the control after 6 and 10 days. Polyphenoloxidase (PPO) activity increased during ripening and was lower in treated fruit than the control. Peroxidase (POD) activity increased during ripening and was higher in treated fruit than the control. SA and MT combination treatment provided no additional positive effects on most parameters. It is concluded that postharvest dipping in 2 mM SA or 0.2 mM MT delayed ripening and improved quality of 'Sensation' mangoes via inhibiting hydrolytic enzymes and enhancing antioxidant system of fruit and are suggested as natural alternative to synthetic chemicals.

Keywords: *Mangifera indica* L., Shelf life, Physicochemical changes, Antioxidant system

Abbreviations: SA—salicylic acid, MT—melatonin, TA—titratable acidity, MSI—membrane stability index, TSS—total soluble solids, TPC—total phenolic content, TFC—total flavonoid content, RSC—radical scavenging capacity, PPO—polyphenoloxidase, POD—peroxidase, Abs—absorbance

INTRODUCTION

Mango fruits (*Mangifera indica* L.) are very popular among consumers due to its high nutritional value, health benefits and distinctive flavor (Sivakumar et al. 2011). Like other climacteric fruits, mango ripening is promoted by ethylene. Following harvesting, pre-climacteric mature hard-green stage fruit ripens rapidly within only a few days at ambient conditions (Mitra and Baldwin 1997; Sivakumar et al. 2011). Accordingly, regulation of fruit ripening and maintenance of quality after harvest using safe natural compounds are alternative approaches to facilitate transportation and marketing (Terry and Joyce 2004; Yashoda et al. 2006; Zhang et al. 2013; Awad et al. 2017; Al-Qurashi and Awad 2018; Liu et al. 2020). In Saudi Arabia, considerable postharvest loss occurs in mangoes including Sensation cultivar, one of the most commercially grown cultivars, due to inappropriate postharvest handling which favors rapid softening during ripening (Awad et al. 2017; Al-Qurashi and Awad 2018). Salicylic acid (SA) is a plant hormone (Raskin 1992) that received attention in postharvest research due to its anti-ripening and anti-browning properties (Shah 2003; Wang and Li 2008). Zeng et al. (2006) found that 1 mM SA vacuum infiltration (−80 kPa) for 2 min plus 10 min at

normal pressure reduced anthracnose incidence and severity in 'Matisu' mangoes inoculated with *Colletotichum gloeosporioides* by activating a number of antioxidant defense enzymes. Pre-storage dipping in 2 mM SA decreased chilling injury incidence and enhanced antioxidant system of 'Zill' mangoes during storage at 5 or 14°C plus 5 days at 25°C (Ding et al. 2007). SA treatment (0.5, 1, 2 or 5 mM) induced resistance against anthracnose and maintained firmness via inhibiting pectin degradation of 'Tainong' mangoes (He et al. 2017). Postharvest dipping in 1.5 or 2.0 mM SA delayed ripening and maintained vitamin C and reduced decay of 'Rangkuai' mangoes at ambient conditions (Mandal et al. 2018). Melatonin (N-acetyl-5-methoxytryptamine, MT) was identified in plants in 1995 (Hattori et al. 1995) and subsequently considered as a phytohormone-like signal molecule possessing several physiological and antioxidant properties (Arnao and Hernández-Ruiz 2015 and 2018). MT affects plant growth and development processes including seed germination, flowering, fruit maturation and ripening as well as stress tolerance (Arnao and Hernández-Ruiz 2015 and 2018; Zhang et al. 2018; Xu et al. 2019; Wang et al.2020; Liu et al. 2020). Postharvest MT treatment (0.05 to 0.5 mM) has been reported to inhibit ethylene, delay ripening and maintain quality of banana fruit (Hu et al. 2017). MT dipping treatment (0.1 mM) down-regulated the expression of PcACS1, PcACO1 and PcPG genes in pears, resulting in delayed ripening and reduced physiological disorders (Zhai et al. 2018). However, to the best of our knowledge, there are only two studies that were very recently published on MT effects on mangoes ripening. In the first study, MT dipping at 1 mM for 3 min maintained quality and antioxidant capacity of mangoes (an unknown cultivar) during 4 weeks at 15°C (Rastegar et al. 2020). The second study showed that MT dipping at 0.5 mM for 1 h modulated ripening of 'Guifei' mangoes at ambient conditions via inhibiting ethylene and ABA biosynthesis (Liu et al. 2020). Therefore, the current study aims to evaluate the response of 'Sensation' mangoes to postharvest dipping in SA at 2 mM or MT at 0.2 mM either alone or in combination as an attempt to regulate ripening and improve quality at ambient conditions.

MATERIALS AND METHODS

Plant Materials and Experimental Procedure

During the growing season of August, 2020, uniform samples of 'Sensation' mangoes were harvested at mature hard-green stage from a commercial orchard in Jizan region (17.4751° N, 42.7076° E), Saudi Arabia. The fruits were packed in perforated carton (20 fruits each) and transported to the postharvest laboratory at King Abdulaziz University in Jeddah within about 5 h at 15°C. Fruit of uniform size, weight (150-200 g/fruit) and color and free of visual defects were selected for this experiment.

Fruit Treatments

Fruit of each treatment/replicate were drenched for 10 min in water (control), salicylic acid (2 mM) or melatonin (0.2 mM) solutions either alone or in combination. A wetting agent, Tween 20 (1 mL L^{-1}), was added in SA and MT solutions. Mangoes were air dried for 1 h, weighed, and stored at 23±1°C and 60–70% (RH) in perforated cartons for 10 days. A separate three replicates (5 fruit each) for each treatment were stored at the same conditions and periodically weighed (at 0, 3, 6, and 10 days) for weight loss expressed in percentage. At the beginning of storage (0 day) and after 3, 6. and 10 days of ripening, additional three samples (5 fruit each) were randomly collected for initial quality and biochemical measurements as detailed below. Following peel color and pulp firmness measurements, samples of both pulp and peel were collected, sliced, and stored at –80°C for total phenol and flavonoid contents, antioxidant capacity, and enzyme activity measurements. Additional portions of fruit pulp were used for TSS, TA, and vitamin C determinations.

Peel Color

Peel color was measured in 5 randomly selected fruits per replicate by using a Minolta Chroma Meter CR-410 (Minolta Camera Co. Ltd., Osaka, Japan). The values of L*, a*, and b* were measured in the middle of each of the five fruit/ replicate. Chroma = $(a^{*2}+b^{*2})^{½}$ represented the hypotenuse of a right triangle with values ranging from 0 = least intense to 60 = most intense. The chroma values indicated the saturation of the color.

Firmness, TSS, TA and Vitamin C Measurements in Fruit Pulp

Pulp firmness of fruit was measured in 5 fruits (two opposite measurements in the middle of each fruit) per replicate using a digital force gauge, model BFG 50N (Mecmesin, Sterling, Virginia, USA) with a probe of 11 mm diameter and the results were expressed in Newtons. A homogeneous sample was prepared from these 5 fruits per replicate for measuring TSS, TA, and vitamin C contents. TSS content was measured in fruit pulp juice with a digital refractometer (Pocket Refractometer PAL 3, ATAGO, Japan) and expressed in percentage. TA was determined in distilled water-diluted fruit juice (1:2) by titrating with 0.1N sodium hydroxide

up to *pH* 8.2 using an automatic titrator (HI 902, HANNA Instrument, USA) and the results were expressed in percentage of citric acid. Vitamin C was measured by titrating the juice sample with freshly prepared dye solution of 2,6-dichlorophenol-indophenol until the color turned pink color and the results were expressed in g kg^{-1} on a fresh weight (FW) basis (Ranganna 2000).

Membrane Stability

Membrane stability was measured in peel disks according to Awad et al. (2017) and was expressed in membrane stability index percentage (MSI %). Three grams of peel disks per replicate/treatment were randomly taken and placed in 30 mL of deionized water at ambient temperature for 4 h in a shaker. Conductivity before boiling (C1) was measured with an electrical conductivity digital meter (Orion 150A+, Thermo Electron Corporation, USA). The same disks were kept in a boiling water bath (100°C) for 30 min to release all electrolytes, cooled to 22±2°C with running water, and conductivity after boiling was recorded (C2). MSI was expressed in percentage using the formula:

$$[1–(C1/C2)] \times 100$$

Preparation of Methanol Extract of Peel and Pulp

TPC and TFC contents, RSC, and enzymes activities were also measured in the fruit peel since the biochemical changes in the peel reflect the ripening and senescence status of the whole fruit including the pulp. In addition, it is well known that the peel contains a much higher level of antioxidant substances and enzymes than the pulp since it represents the first line of defense against biotic and abiotic stresses that ultimately affect fruit quality and ripening attributes. Two grams of fruit peel and pulp (randomly collected from 5 fruits/replicate) were extracted by shaking at 150 rpm for 12 h with 20 ml methanol (80%) and filtered with Whatman No. 1 paper. The filtrate designated as methanol extract was used for total phenols, total flavonoids, and antioxidant activity estimations.

Estimation of Total Phenol and Flavonoid Contents

TPC was measured according to Hoff and Singleton (1977). Fifty µL of the methanol extract was mixed with 100 µL Folin - Ciocalteu reagent, 850 µL of methanol, and allowed to stand for 5 min at ambient temperature. Five hundred µL of 20% sodium carbonate was added and allowed to react for 30 min. Absorbance was measured at 750 nm. TPC was quantified from a calibration curve obtained by measuring the absorbance of known concentrations of gallic acid and the results were expressed as g kg^{-1} FW gallic acid equivalent. TFC was determined using a modified colorimetric method described previously by Zhishen et al. (1999). Methanol extract or standard solution (250 µL) was mixed with distilled water (1.25 mL) and 5 % $NaNO_2$ solution (75 µL). After standing for 6 min, the mixture was combined with 10% $AlCl_3$ solution (150 µL), 1 M NaOH (0.5 mL) and distilled water (275 µL) were added to the mixture 5 min later. The absorbance of the solutions at 510 nm was then measured. TFC was quantified from a calibration curve obtained by measuring the absorbance of known concentrations of catechin and the results expressed as g kg^{-1} FW catechin equivalent.

Evaluation of DPPH Radical Scavenging Capacity of Peel and Pulp

Free radical scavenging activity of methanol extract of peel and pulp was determined using the 1,1-diphenyl-2-picrylhydrazyl (DPPH) method (Ao et al. 2008). A methanol extract (0.1 mL) was added to 0.9 mL of freshly prepared DPPH methanol solution (0.1 *m*M). An equal amount of methanol was used as a control. After incubation for 30 min at room temperature in the dark, the absorbance (Abs) was measured at 517 nm using a spectrophotometer. Scavenging activity (%) was calculated using the following formula:

$$\text{DPPH radical scavenging \%} = [(\text{Abs control} – \text{Abs sample})/\text{Abs control}] \times 100$$

The inhibition concentration (IC_{50}) was defined as µg phenolics of the test sample that decreases 50% of initial radical. The IC_{50} values were calculated from the dose responses curves.

Enzymes Measurements of Peel and Pulp

Crude Extract

One gram of fruit peel and pulp (randomly collected from 5 fruit/ replicate) was homogenized with 20 mM Tris–HCl buffer, *pH* 7.2 using homogenizer. The homogenate was centrifuged at 10,000 rpm for 10 min at 4°C. The supernatant was designed as crude extract and stored at -20°C for peroxidase, polyphenoloxidase, polygalacturonase, xylanase, and α-amylase assays.

Polyphenoloxidase Assay

PPO (EC 1.14.18.1) activity was assayed with catechol as a substrate according to the spectrophotometric procedure of Jiang et al. (2002). The crude extract (0.2 mL) was rapidly added to 2.8 mL of 20 mM catechol solution prepared in 0.01 M sodium phosphate buffer (*pH* 6.8). The increase in absorbance at 400 nm was recorded for 3 min using a spectrophotometer. One unit of enzyme

activity was defined as the amount of the enzyme that causes a change of 0.1 in absorbance per min under standard assay conditions.

Peroxidase Assay

POD (EC 1.11.1.7) activity was assayed according to Miranda et al. (1995). The reaction mixture in one mL contained the following: 8 mM H_2O_2, 40 mM guaiacol, 50 mM sodium acetate buffer, pH 5.5 and 0.1 mL crude extract. The change in absorbance at 470 nm due to guaiacol oxidation was followed for 1 min using a spectrophotometer. One unit of peroxidase activity was defined as the amount of enzyme that increases the O.D. by 1.0 per min under standard assay conditions.

Polygalacturonase, α-amylase and Xylanase Assays

PG (EC 3.2.1.15), α-amylase (EC 3.2.1.1) and xylanase (EC 3.2.1.8) activities were assayed by determining the liberated reducing end products using galacturonic acid, maltose and xylose, respectively as standards (Miller 1959). The reaction mixture (0.5 mL) contained 5 mg substrate, 0.25 mL of 0.2 M sodium acetate buffer, pH 5.5 and 50 µL of crude extract. Assays were carried out at 37°C for 1 h. Then 0.5 mL dinitrosalicylic acid reagent was added to each tube and heated in a boiling water bath for 10 min. After cooling to room temperature, the absorbance was measured at 560 nm. Substrates used were polygalacturonic acid, starch and xylane for PG, α-amylase and xylanase, respectively. One unit of enzyme activity was defined as the amount of enzyme which liberated 1 µM of reducing sugar per min under standard assay conditions.

Statistical Analysis

The experiment was laid in a Completely Randomized Design with three replicates per treatment. Analysis of variance (ANOVA) was done using the statistical package software SAS (SAS Institute Inc., 2000, Cary, NC., USA). Comparisons between means were made by the Least Significant Difference (LSD) at $P \leq 5\%$.

RESULTS AND DISCUSSION

Fruit quality is determined both by physical (especially firmness) and biochemical parameters including pigments (color), TSS, TA, and TSS/TA ratio as well as antioxidants content that contribute to their health value and marketability. As climacteric fruit, mango exhibits high metabolic activities leading to rapid softening associated with an increase in TSS and a decrease in TA during ripening at ambient conditions (Sivakumar et al. 2011). Thus, the use of natural compounds such as SA and MT to regulate fruit ripening and reduce postharvest losses are explored. In the current study, there were significant interaction effects between treatments and ripening periods on all of the parameters, thus we passed over the main effects and focused instead on the interactions. In addition, the combination treatment of SA and MT did not result in significant difference in most of the parameters compared to SA or MT treatment alone.

Peel Color

Peel color is an important quality parameter that influences mango fruit marketability. The color parameters L*, a*, b*, and chroma values of peel increased during ripening and were significantly lower in MT- and SA-treated fruit than the control (Fig. 1). The increase in L*, b*, and chroma values during ripening indicates an increase in lightness, yellowness and vividness in color of fruit peel, respectively. The gradual increase of a* value during ripening reflects chlorophyll degradation possibly by the action of chlorophyllase, as well as to the accumulation of carotenoid and xanthophyll pigments which lead to decrease of greenness and/or increasing redness (Baez-Sanudo et al. 2009). Rastegar et al. (2020) recorded an increase in a* value of mango fruit peel during cold storage but with no significant effects for MT treatment compared to the control. In contrast, MT treatment slowed the changes in L* and a* values and delayed senescence of litchis (Zhang et al. 2018) and increased L* values of strawberries during cold storage

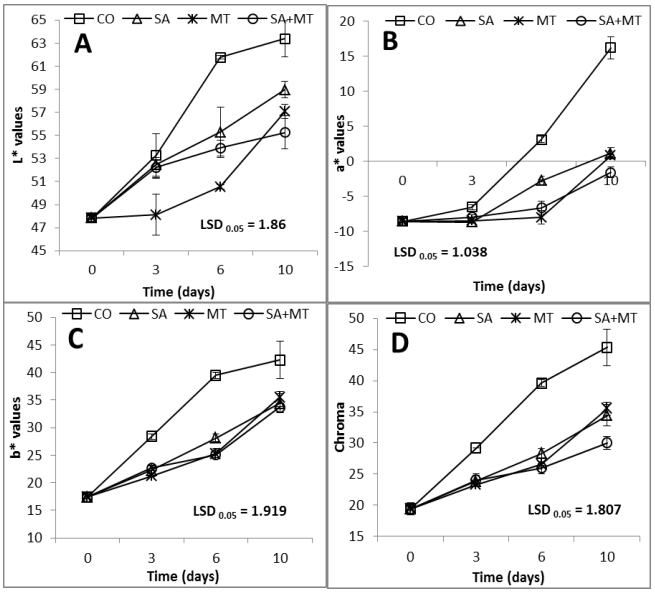

Fig. 1. The interaction effects between treatments (2 mM salicylic acid (SA), 0.2 mM melatonin (MT) alone or in combination (SA+MT) and control (CO)) and ripening period on peel color parameters L* (A), a* (B), b* values (C) and chroma (D) of 'Sensation' mangoes during 10 days of ripening at 23±1°C and 60–70% RH. Values are means ± SD from three replicates (n = 3). Statistical analysis was performed using LSD test at $P \leq 5\%$.

compared to the control (Liu et al. 2018). The results with postharvest SA dipping at 2 mM conformed with that of 'Flordaking' peaches wherein higher skin luminosity and lower a* values than control were obtained during cold storage (Tareen et al. 2012). In addition, SA-treated strawberries showed lower a* values than the control during cold storage (Shafiee et al. 2010).

Weight Loss

Weight loss gradually increased during ripening reaching 4.48% in the control after 10 days and was significantly lower in treated fruit than the control particularly after 6 and 10 days. The lowest weight loss (3.88%) was obtained in MT-treated fruit (Fig. 2). Weight loss is a critical quality parameter that reflects freshness of fruit. It is well known that both transpiration and respiration contribute to fruit weight loss during ripening (Narayana et al. 1996; Razzaq et al. 2015). Our results validated those of He et al. (2017) and Mandal et al. (2018) where SA treatment reduced weight loss during ripening at ambient conditions. Regarding MT effects, our results are in agreement with those of Rastegar et al. (2020) on an unknown mango cultivar during cold storage, and Liu et al. (2020) on 'Guifei' mangoes during ripening at ambient conditions.

Firmness, MSI, and Hydrolytic Enzymes

Firmness sharply decreased during ripening and was higher in treated fruit than the control especially after 3 and 6 days. After 6 days, fruit firmness was almost double in treated fruit compared to the control. However, after 10 days, only MT-treated fruit retained significantly higher firmness (7.5 N) than the control (4.4 N) (Fig. 2). MSI of fruit peel sharply decreased during ripening and was significantly higher in all treatments than the control (Fig. 2). Firmness is also a critical quality factor that determines mango fruit ripening, storability, transportation and marketability (Jha et al. 2010; Sivakumar et al. 2011). SA and MT treatments slowed down fruit softening possibly by suppression of ethylene production. SA treatment has been shown to delay ripening and to suppress ethylene production and respiration rates during ripening at ambient conditions in four mango cultivars including 'Sensation' (Prasad and Sharma 2018), 'Zill' (Hong et al. 2014) and 'Chausa' during cold storage (Barman and Asrey 2014). MT dipping at 0.5 mM for 1 h delayed 'Guifei' mangoes ripening during 10 days at ambient conditions via inhibiting ethylene and ABA biosynthesis (Liu et al. 2020). However, Rastegar et al. (2020) found that 1 mM MT dipping of mangoes retained higher firmness, vitamin C and TPC contents, and antioxidant activity, but with no impact on TSS and TA contents during 4 weeks of storage at 15°C.

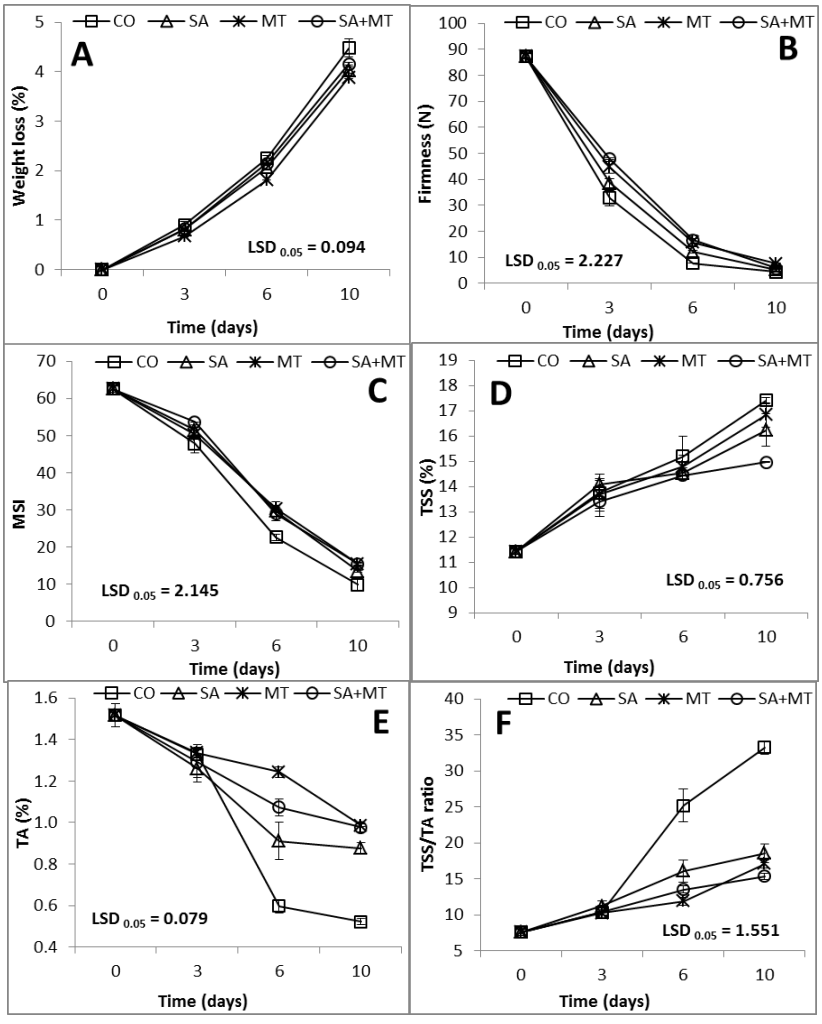

Fig. 2. The interaction effects between treatments (2 mM salicylic acid (SA), 0.2 mM melatonin (MT) alone or in combination (SA+MT) and control (CO)) and ripening period on weight loss (A), firmness (B), membrane stability index (MSI) (C), total soluble solids (TSS) (D), titratable acidity (TA) (E) and TSS/TA ratio (F) of 'Sensation' mangoes during 10 days of ripening at 23±1°C and 60–70% RH. Values are means ± SD from three replicates (n = 3). Statistical analysis was performed using LSD test at *P* ≤ 5%.

PG activity in peel was higher after 3 and 6 days than initial in the SA and MT combination treatment and the control followed by a sharp decrease after 10 days (Fig. 3a). MT-treated fruit exhibited

higher PG activity than initial only after 6 days but sharply decreased thereafter. The treated fruit showed lower PG activity than the control after 3 and 6 days. On the other hand, PG activity in pulp increased particularly in the control after 3 and 6 days, while SA treatment was similar to the initial value (Fig. 3b). However, after 10 days, PG activity in both peel and pulp sharply decreased in all treatments, except for the control that remained constant. Xylanase activity in peel increased after 3 and 6 days, except for MT after 3 days, and SA combined with MT that remained constant (Fig. 3c). In the pulp, xylanase activity similarly increased after 3 and 6 days, except for SA alone or combined with MT after 3 days, and MT alone or combined with SA treatments after 6 days, that remained constant (Fig. 3d). However, after 10 days, xylanase activity in both peel and pulp sharply decreased in all treatments during ripening and was significantly lower in treated fruit than control. During ripening on the 3rd and 6th day, α-amylase activity of peel and pulp increased in all treatments (Fig. 3e) except for the pulp of fruits subjected to MT alone or combined with SA treatments (Fig. 3f). However, after 10 days, α-amylase activity sharply decreased in all treatments in both peel and pulp and was significantly lower in treated fruit than control.

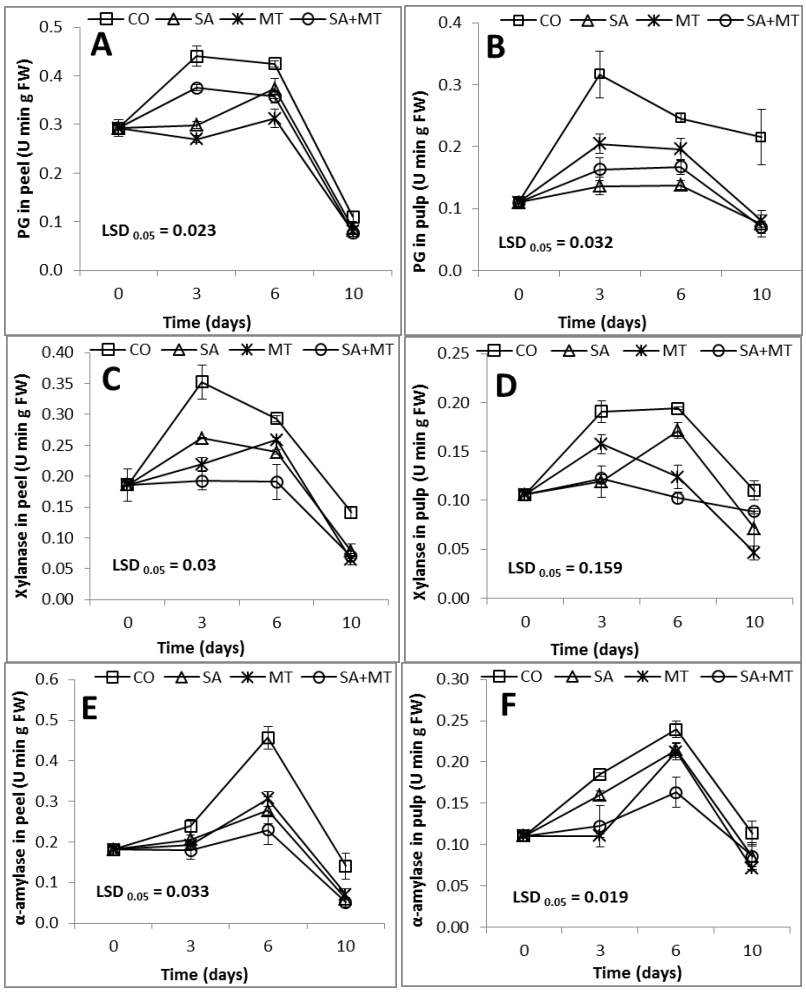

Fig. 3. The interaction effects between treatments (2 mM salicylic acid (SA), 0.2 mM melatonin (MT) alone or in combination (SA+MT) and control (CO)) and ripening period on activities of polygalacturornase (PG) in peel (A) and pulp (B), xylanase in peel (C) and pulp (D) and α-amylase in peel (E) and pulp (F) of 'Sensation' mangoes during 10 days of ripening at 23±1°C and 60–70% RH. Values are means ± SD from three replicates (n = 3). Statistical analysis was performed using LSD test at $P \leq 5\%$.

The rapid decrease in fruit firmness and MSI during ripening is possibly due to disassembly and/or depolymerization of the cell wall pectin matrix and hemicelluloses by hydrolytic enzyme action under the control of ethylene and ABA (Brummell 2006; Cao et al. 2018; Xiao et al. 2019). In the current study, PG, xylanase, and α-amylase activities in both peel and pulp were higher than initial but were much lower in MT or SA treated fruit than in the control. Previous studies indicated that MT inhibited PG, pectin methylesterase (PME), and β-galactosidase (β-Gal) activities and reduced pectin and polysaccharides solubilization and depolymerization via inhibition of ethylene and ABA biosynthesis in 'Guifei' mango during ripening (Liu et al. 2020). It was reported that MT treatment delayed textural changes and cell wall degradation of three pear cultivars by inhibiting ethylene production, PG, and cellulase gene expression (Zhai et al. 2018) and maintained firmness of stored peaches via down-regulations of genes associated with cell wall disassembly (Gao et al. 2016). SA retained firmness in other mango cultivars via inhibiting the conversion of insoluble protopectin into soluble pectin (He et al. 2017) and lowering PG and PME activities during ripening at 25°C (Barman and Asrey 2014).

TSS, TA and TSS/TA Ratio

TSS increased during ripening and was significantly lower in SA- and MT-treated fruit than the control only after 10 days (Fig. 2d). SA in combination with MT treatment resulted in lower TSS than the other treatments including the control. TA decreased during ripening and was significantly higher in treated fruit after

6 and 10 days than the control (Fig. 2e). TSS/TA ratio significantly increased during ripening and was lower in treated fruit, after 6 and 10 days than the control (Fig. 2f). The combination treatment of MT and SA had lower TSS/TA ratio than other treatments including control after 6 and 10 days.

The observed changes in TSS might be attributed to the reduction in *α*-amylase activity during ripening by MT and SA treatments. The increase in TSS and TSS/TA ratio and the decrease in TA reflects mango ripening advancement and the conversion of starch into simple sugars mainly by the action of hydrolytic enzymes (Kittur et al. 2001). MT retarded the increase in TSS, and retained TA of 'Guifei' mangoes during ripening at ambient conditions (Liu et al. 2020). On the other hand, Rastegar et al. (2020) reported that TSS increased and TA decreased during cold storage but with no significant effect of MT on an unknown mango cultivar. In addition, MT treatment showed no significant effect on TSS and TA contents of strawberries during storage (Liu et al. 2018).

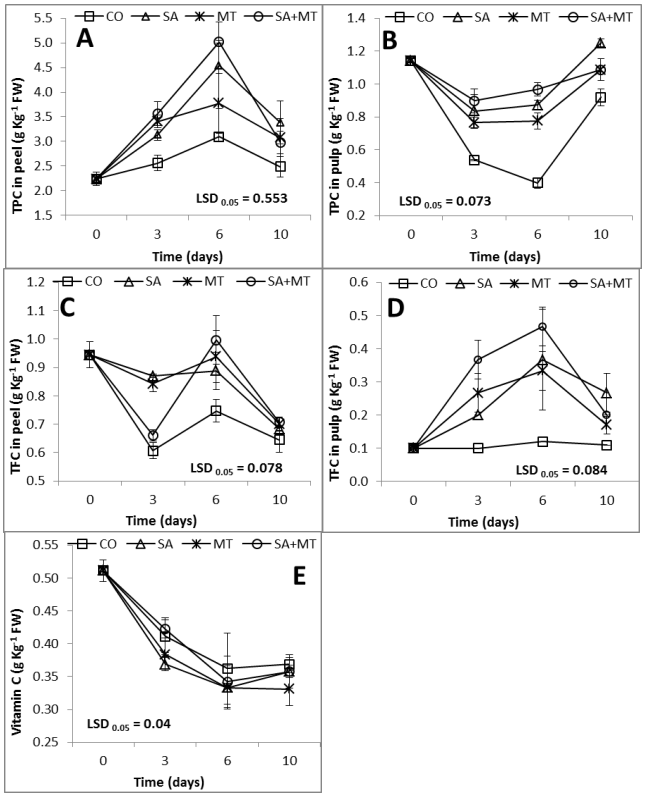

Fig. 4. The interaction effects between treatments (2 *m*M salicylic acid (SA), 0.2 *m*M melatonin (MT) alone or in combination (SA+MT) and control (CO)) and ripening period on total phenol (TPC) in peel (A) and pulp (B), total flavonoid (TFC) in peel (C) and pulp (D) and vitamin C in pulp (E) contents of 'Sensation' mangoes during 10 days of ripening at 23±1°C and 60–70% RH. Values are means ± SD from three replicates (n = 3). Statistical analysis was performed using LSD test at *P* ≤ 5%.

Our results confirmed those of Mandal et al. (2018) where SA-treated 'Rangkuai' mangoes exhibited lower TSS and TSS/TA ratio and higher TA than the control during 15 days of ripening at ambient conditions. Also, SA treatment maintained TSS and delayed the decrease in TA in 'Zill' mangoes (Hong et al. 2014) and in 'Chausa' mangoes during cold storage (Barman and Asrey 2014).

Antioxidant Compounds and Activity

TPC in peel increased after 3 and 6 days during ripening and was higher in treated fruit than the control, but decreased thereafter (Fig. 4a). In the pulp, TPC decreased after 3 and 6 days, and was higher in treated fruit than the control, but increased thereafter (Fig. 4b). TFC in the peel initially decreased followed by fluctuating trends although treated fruit retained higher level than the control after 3 and 6 days (Fig. 4c). In the pulp, TFC increased followed by fluctuating trends as in the peel in the case of treated fruit, and values were higher than the control particularly after 3 and 6 days (Fig. 4d). SA and MT combination treatment exhibited higher TFC than other treatments including the control after 3 and 6 days. Vitamin C content decreased during ripening and was not affected by the treatments (Fig. 4e). RSC in peel decreased (higher DPPH IC_{50} values) during ripening and was higher (lower DPPH IC_{50} values) in treated fruit than the control (Fig. 5a). However, in the pulp, RSC increased during ripening and was not affected by treatments, except for SA and MT combination that gave higher RSC than the control after 6 and 10 days (Fig. 5b).

Mangoes are important sources of antioxidants including phenolics, flavonoids, carotenoids and vitamins (Sivakumar et al. 2011). Such antioxidants protect fruit against oxidative stress and pathogen attacks as well as their levels determine fruit health value. However, fruit ripening and senescence are considered as oxidative processes in which the transition from maturation to

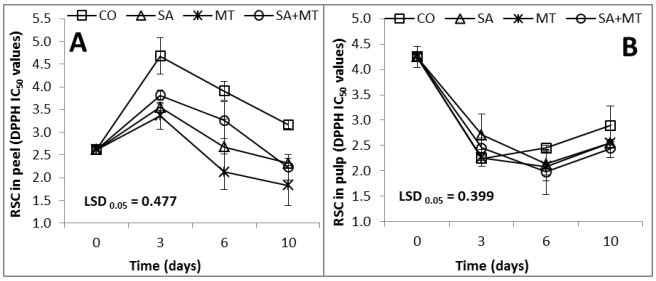

Fig. 5. The interaction effects between treatments (2 *m*M salicylic acid (SA), 0.2 *m*M melatonin (MT) alone or in combination (SA+MT) and control (CO)) and ripening period on DPPH radical scavenging capacity (RSC) in peel (A) and pulp (B) of 'Sensation' mangoes during 10 days of ripening at 23±1°C and 60–70% RH. Values are means ± SD from three replicates (n = 3). Statistical analysis was performed using LSD test at *P* ≤ 5%.

ripening/senescence is accompanied by a progressive shift toward an oxidative state due to antioxidant system reduction (Goulao and Oliveira 2008). The patterns of changes in TPC and TFC especially in mango fruit pulp as well as the positive effects of MT treatment in retaining higher antioxidant compounds than the control are in conformity with those of Rastegar et al. (2020) on other mango cultivars and of Zhang et al. (2018) on litchis. SA treatment has been shown to increase and/or retain TPC and TFC in four mango cultivars including 'Sensation' during normal ripening at ambient conditions (Prasad and Sharma 2018) and in 'Chausa' mangoes during cold storage followed by 3 days of shelf life (Barman and Asrey 2014). Our results on vitamin C content partially contradicted with those of Junmatong et al. (2015) and (Mandal et al. 2018) in which SA treatment of other mango cultivars retained higher vitamin C than the control during ripening at ambient conditions. Also, MT-treated mangoes at higher dose (1 mM) retained higher vitamin C than the control during cold storage (Rastegar et al. 2020). Our results on RSC partially contradict with those of Rastegar et al. (2020) in which RSC decreased in mango fruit pulp treated or not with MT during cold storage, but at the end of storage MT treated fruit showed higher RSC than control.

PPO and POD

PPO activity in both peel and pulp greatly increased during ripening and was significantly lower in treated fruit than the control (Fig. 6a, b). MT-treated fruit showed the lowest PPO activity in the peel after 3 and 6 days. POD activity in fruit peel increased up to 3 days in all treatments including the control (Fig. 6c). After 6 and 10 days, POD activity gradually decreased in the control fruit with no significant changes in the treated fruit, except for SA treatment that showed higher POD. However, in the pulp, POD activity increased with fluctuations during ripening in all treatments, except for the control that increased up to 3 days then remained constant thereafter (Fig. 6d). The treated fruit exhibited much higher POD activity than control during all ripening periods, except for SA treatment after 3 days that showed similar level as that of the control.

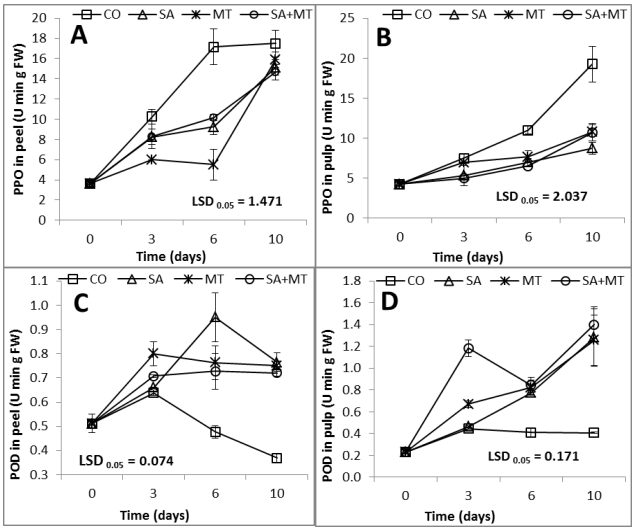

Fig. 6. The interaction effects between treatments (2 mM salicylic acid (SA), 0.2 mM melatonin (MT) alone or in combination (SA+MT) and control (CO)) and ripening period on activities of polyphenoloxidase (PPO) in peel (A) and pulp (B) and peroxidase (POD) in peel (C) and pulp (D) of 'Sensation' mangoes during 10 days of ripening at 23±1°C and 60–70% RH. Values are means ± SD from three replicates (n = 3). Statistical analysis was performed using LSD test at *P* ≤ 5%.

Rastegar et al. (2020) reported that PPO in mango fruit pulp significantly increased during storage and was lower in MT-treated fruit during cold storage. PPO is generally associated with tissue browning of fresh horticultural commodities via oxidizing phenolic compounds into quinones during ripening and senescence (Yoruk and Marshall 2003). A similar pattern of POD change in the pulp was reported by Rastegar et al. (2020) in which MT-treated mangoes showed higher POD activity than the control during cold storage. It has been reported that 0.1 MT produced contradicting effects on POD activity in peaches during storage where MT decreased POD activity at 1°C, in contrast to the results at room temperature (Gao et al. 2016 and 2018). On the other hand, MT treatment had no significant effects on POD activity in pear fruit during ripening at 26°C (Zhai et al. 2018). Regarding SA effects, our results confirmed those of Ding et al. (2007) and of Vishwakarma et al. (2018) where SA treatment increased the antioxidant enzyme activities including POD but decreased PPO activity of mango fruit during ripening. The lack of additive effects for the SA and MT combination treatment on fruit quality during ripening compared to when SA or MT was used alone suggests a need for further study.

CONCLUSION

Postharvest dipping in 2 mM SA or 0.2 mM MT delayed ripening, maintained and improved quality of 'Sensation' mangoes at ambient conditions via inhibiting hydrolytic enzymes and enhancing antioxidant system of fruit and is being suggested as natural alternative to synthetic chemicals. However, SA and MT combination treatment provided no additional positive effects on most quality parameters of fruit.

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